Radiometric analysis of blue grenadier, Macruronus novaezelandiae, otolith cores

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Blue grenadier, Macruronus novaezelandiae, is a major commercial fish species in the upper continental slope waters of southeastern Australia and New Zealand, An accurate knowledge of age and growth rate of this species is required for management of the fishery. Several ageing studies have provided estimates of age for blue grenadier (hoki) from New Zealand waters (reviewed by Paul, 1992), with the maximum age ranging from 12 to 15 years. Kenchington and Augustine (1987) examined whole otoliths and thin transverse sections from blue grenadier caught in southeastern Australian waters and reported a maximum age of 25 years. However. Kenchington and Augustine (1987) were unable to validate their ages for fish >3 years of age. They found that conventional techniques of validating age estimates, such as tagging, modal analysis, marginal increment or edge-type analysis, and back calculation of lengths at age, could not be applied to blue grenadier.

The possibility that radiometric analysis of fish otoliths, a technique pioneered by Bennett et al. (1982) for ageing fish, might validate the ages of blue grenadier led to a study by Fenton et al. (1990). Unfortunately radiometric analysis of ²¹⁰Pb and ²²⁶Ra in whole blue grenadier otoliths was unsuccessful for estimating fish ages (Fenton et al., 1990). This was due to an exponential reduction in ²²⁶Ra incorporation into the otoliths of blue grenadier during their life, a reduction that was believed to result from an ontogenetic change in habitat from juvenile to adult fish.

Recent advances with radiometric ageing of fish indicate that ages can be determined radiometrically from otolith cores (Campana et al., 1990, 1993; Smith et al., 1991). The coring method removes all the layers of otolith growth beyond the earliest year(s) and thus avoids problems of changes in ²²⁶Ra or ²¹⁰Pb uptake patterns in later life. Furthermore, an analysis of otolith cores circumvents the need to model the mass growth rate of otoliths, which is necessary in determining age from whole otoliths (Smith et al., 1991).

Radiometric analysis of otolith cores has been successfully used to age redfish, *Sebastes mentella* (Campana et al., 1990), and splitnose rockfish, *Sebastes diploproa* (Smith et al., 1991), with ²¹⁰Pb/²²⁶

Ra. In addition, otolith cores have been analyzed with ²²⁸Th/²²⁸Ra to age flying fish, *Hirundichthys affinis* (Smith et al., 1991; Campana et al., 1993), and silver hake, *Merluccis bilinearis* (Smith et al., 1991). While the isotope pair ²²⁸Th/²²⁸Ra is useful for short-lived fish up to about 5 years (Campana et al., 1993), analysis of ²¹⁰Pb/²²⁶Ra is appropriate for medium-aged to long-lived fish up to about 120 years.

In the present study measurements of ²¹⁰Pb/²²⁶Ra disequilibria have been conducted on cores of adult female blue grenadier otoliths in an attempt to provide independent age data for this species.

Materials and methods

Field collection

Blue grenadier, Macruronus novaezelandiae, were collected by bottom trawling (500–850 m depth) during cruises conducted northwest of Sandy Cape off the west Coast of Tasmania by the Tasmanian Department Primary Industry, Division of Sea Fisheries, and the CSIRO Division of Fisheries between 1985 and 1986. Sagittal otoliths were removed and stored dry. This collection of otoliths was used by Kenchington and Augustine to estimate age by annulus counts (1987) and was subsequently made available for the present study.

Otolith morphometrics

The linear dimensions of blue grenadier otoliths (maximum length, width, and thickness with respect to the primordia) were measured to the nearest 0.1 mm with calipers. Otolith weight was also determined for n=172 blue grenadier otoliths

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from fish previously aged by Kenchington and Augustine (1987). The dimensions of otoliths from 1+ fish (ages assigned by Kenchington and Augustine, 1987) were examined and the average linear dimensions and weight for a 1+ fish determined. The average otolith dimensions of 1+ fish were selected as the shape of the core to be removed. This core size was chosen 1) because it ensured minimal influence of changing ²²⁶Ra (as reported in Fenton et al., 1990); 2) because it represented the minimum core size that could be repeatably isolated (attempts to remove smaller cores, e.g. 0+ dimensions, resulted in the otolith shattering or splitting, and being unusable; and 3) because it fitted the age estimate of 1+ otoliths by Kenchington and Augustine (1987) who were able with confidence to validate fish age for individuals under 3 years old.

Removal of otolith core

Blue grenadier otoliths chosen for coring were selected from females collected from the same site, on the same date, and of similar fish length and otolith weight. Cores were removed from the otoliths by sanding the outer layers away by hand (with respect to the primordia) with wet and dry sand-paper (silicon carbide paper) in progressively finer grades until the desired core dimensions were achieved. The mean core dimensions were compared with the mean whole otolith dimensions for 1+, 2+, and 3+ fish. New, wet and dry sand paper was used for each otolith to eliminate the risk of ²¹⁰Po cross contamination between otoliths. The cores of 2 or 3 fish were pooled to produce the 1-g sample necessary for radiometric analysis.

Radiometric analysis

Cores were cleaned of adventitious ²¹⁰Po by exposure to an alkaline H₂O₂ solution for 1/2 hour. The analysis of ²¹⁰Pb via its alpha-emitting, short-lived daughter proxy ²¹⁰Po followed the method we have previously described (Fenton et al., 1991), employing high resolution alpha-spectrometry. Polonium-210 was assumed to be in equilibrium with ²¹⁰Pb in all samples, and ²¹⁰Pb concentrations were corrected back to the date of fish collection. Mean ²¹⁰Po reagent blank was 0.0071 ±0.0012 disintergrations per minute (dpm·g⁻¹). Recovery of ²¹⁰Po was invariably >90%. Instrumental background counts (for ²⁰⁸Po and ²¹⁰Po) were less than 1 count per day. Analysis of ²²⁶Ra was by a direct alpha spectrometry technique (Fenton et al., 1991). The mean activity of the ²²⁶Ra blanks was 0.0174 ±0.002 dpm·g-1. Both 210Po and 226Ra blank values were reduced to lower values than those from previous studies because of tighter quality control of reagents and minor improvements in technique.

Stable element analysis

The levels of lead (Pb) and barium (Ba) are presumed to act as stable equivalents of ²¹⁰Pb and ²²⁶Ra (Fenton and Short, 1992) and, as such, can potentially be used to assess the uptake of the radioactive isotopes and for normalizing the radiometric data. Therefore the concentrations of stable lead, barium, calcium (Ca), and strontium (Sr) in each otolith core sample were measured. An aliquot of the same dissolved otolith core solution used for ²¹⁰Pb and ²²⁶Ra analysis was analyzed. Lead and barium were analyzed by inductively coupled plasma mass spectrometry (ICP-MS) and strontium and calcium by inductively coupled plasma atomic emission spectrometry (ICP-AES).

Calculation of fish age

Fish age was calculated by using the model proposed by Campana et al. (1990) and described in Smith et al. (1991). The ²¹⁰Pb/²²⁶Ra activity ratio of the otolith core during growth, assuming a constant mass growth model for the period of core formation only, is given by the equation:

$$\frac{A_{Pb^{210}}}{A_{Ra^{226}}} = 1 - (1 - R) \left(\frac{1 - e^{-\lambda T}}{\lambda T} \right),$$

where A = specific activity (i.e. activity concentration)

R = initial activity ratio (i.e. uptake activity
ratio):

 $\lambda = \text{decay constant of }^{210}\text{Pb (year}^{-1)};$

T = period of core formation (years).

The activity ratio of the core at any subsequent time in the life of the fish is unaffected by mass growth of the otolith and is therefore given by the equation:

$$\frac{A_{Pb^{210}}}{A_{Ra^{226}}} = \left(1 - e^{-\lambda(t-T)}\right) + \left[1 - (1-R)\left(\frac{1 - e^{-\lambda T}}{\lambda T}\right)\right]e^{-\lambda(t-T)},$$

where t= age of the fish (years).

The fish age is found by solving for t by the Newton-Raphson iterative method. All radiometric age values are given with an error value of ± 1 standard deviation.

Table 1

Comparison of core dimensions and the otolith dimensions of 0+, 1+, 2+, and 3+ blue grenadier, *Macruronus novaezelandiae*. Probability (P) values are given for t-test comparison of the mean core dimensions and mean whole otolith dimensions SD= standard deviation and n is the number of otoliths measured.

	Core dimension	0+ otolith	1+otolith	2+ otolith	3+ otolitl
Otolith length (mm)					
Mean	12.50	7.20	12.26	15.94	18.66
SD	0.20	0.29	0.68	1.37	1.09
n	24	48	38	12	14
P		< 0.001	>0.02	<0.001	<0.001
Otolith width (mm)					
Mean	5.44	3.27	5.40	6.73	7.64
SD	0.14	0.15	0.30	0.69	0.33
n	24	47	38	12	14
P		<0.001	> 0.50	< 0.001	<0.00
Otolith Thickness (mm)					
Mean	1.28	0.74	1.25	1.69	1.79
SD	0.05	0.07	0.10	0.04	0.14
n	24	46	38	12	14
P		<0.001	> 0.10	<0.001	<0.00
Otolith Weight (g)					
Mean	0.2008	0.0197	0.0961	0.1870	0.275
SD	0.007	0.020	0.013	0.043	0.030
n	24	57	38	13	14
P		<<0.001	<<0.001	0.50-0.20	<<0.00

Results

Otolith core dimensions

Otolith growth in linear dimensions and weight for juvenile blue grenadier (0+, 1+, 2+, and 3+) was rapid (Table 1). The cores that were removed approximated the linear dimensions of 1+ fish, but they were heavier than the average 1+ fish with a weight that was not significantly different from that of 2+ fish (t=1.15;P>0.20). The dimensions and weight of 3+ fish otoliths were significantly greater than the isolated core dimensions. Therefore, in view of the core weight our calculation of age assumed that the cores analyzed represented 2 years of growth.

Stable element analysis

Low levels of both stable lead and barium were found in all the samples (Table 2). Stable lead ranged from 0.269 to 1.78 ppm. Barium levels were low, ranging from 1.50 to 2.28 ppm, consistent with the relatively low ²²⁶Ra levels in these blue grenadier otolith cores. The ratio of Pb/Ba was relatively high, ranging from 0.17 to 0.78. From this Pb/Ba data we concluded that

there is some allogenic uptake of ²¹⁰Pb from the environment. However the extent of uptake was difficult to assess given the low levels of Pb and Ba measured. The Sr/Ca ratio ranged between 0.0034 and 0.0042.

Radiometric results

²¹⁰Pb ranged from 0.0040 ± 0.0045 to 0.0078 ± 0.0025 dpm·g⁻¹ (Table 3). The ²²⁶Ra content of the otolith

Table 2

Blue grenadier, *Macruronus novaezelandiae*, stable element data from inductively coupled plasma mass spectrometry (ICP-MS) and inductively coupled plasma atomic emission spectrometry (ICP-AES) analysis of otolith core solutions.

Sample number	Pb ppm	Ba ppm	Pb/Ba ppm/ppm	Sr ppm	Ca ppm	Sr/Ca ppm/ppm
LH 2269	0.749	1.50	0.50	1590	400300	0.0040
LH 2270	1.78	2.28	0.78	1620	387800	0.0042
LH 2271	0.389	1.57	0.22	1810	448380	0.0040
LH 2272	0.269	1.55	0.17	1920	483570	0.0040

Table 3

Blue Grenadier, *Macruronus novaezelandiae*, otolith core radiometric results and ages calculated by using individual ²²⁶Ra and mean ²²⁶Ra activity ratios.

Sample number	Mean fish length (cm)	Mean otolith mass (g)	No. of otoliths	²¹⁰ Pb (dpm· g ⁻¹)	²²⁶ Ra (dpm· g ⁻¹)	²¹⁰ Pb/ ²²⁶ Ra activity ratio from individual ²²⁶ Ra activity ratio	²¹⁰ Pb/ ²²⁶ Ra activity ratio from mean ²²⁶ Ra activity ratio	Otolith core age (years) from individual ²²⁶ Ra activity ratio	Otolith core age (years) from mean ²²⁶ Ra activity ratio
LH 2269	95.63 ±0.44	0.5395 ±0.009	6	0.0051 ±0.0023	0.0290 ±0.0036	0.174 ±0.082	0.227 ±0.111	5.5 (+3.4, -3.0)	7.6 (+5.0, -4.3)
LH 1817	99.3 ±0.52	0.7116 ±0.078	6	0.0040 ±0.0045	0.0203 ±0.0042	0.197 ±0.225	0.179 ±0.205	6.4 (+10.6, -7.9)	5.7 (+9.2, -7.2)
LH 2270	104.0 ±1.79	0.7136 ±0.053	6	0.0078 ±0.0025	0.0179 ±0.0026	0.438 ±0.153	0.351 ±0.129	17.9 (+10.2, -7.7)	13.3 (+7.1, -5.8)
LH 2271	107.2 ±3.06	0.8896 ±0.073	6	0.0074 ±0.0031	0.0222 ±0.0039	0.332 ±0.151	0.331 ±0.151	12.3 (+8.2, -6.5)	12.2 (+8.2, -6.5)
LH 2272	110.3 ±1.37	0.9531 ±0.076	6	0.0058 ±0.0030	0.0223 ±0.0039	0.259 ±0.142	0.259 ±0.143	9.0 (+6.8, -5.6)	9.0 (+6.9, -5.7)

cores ranged from 0.0179 to 0.0290 dpm·g⁻¹ and showed a mean value of 0.0223 ± 0.0036 dpm·g⁻¹. The ²²⁶Ra values found for the cores were consistent with our earlier analysis (Fenton et al., 1990) of whole otoliths (see Discussion section) and further confirmed that the core represents approximately a 2+ fish.

In order to select the value of R, the initial activity ratio used in the calculation of age, we examined our previous data for whole otoliths (Fenton et al., 1990) and the stable element analysis of Pb and Ba (discussed above). From the data for a 1+ fish a value of R < 0.06 is indicated; however, data for 0+ fish indicate R could be as high as 0.1. A value of R = 0.05 appears to be the best estimate for the initial activity. If R were as high as 0.1, it would lower the age estimate by approximately 9 months. Conversely, if R = 0, ages would be higher by 2.6 years. The difference that a lower or higher value makes to the age estimates is well within the error associated with each radiometric age calculated by using R = 0.05.

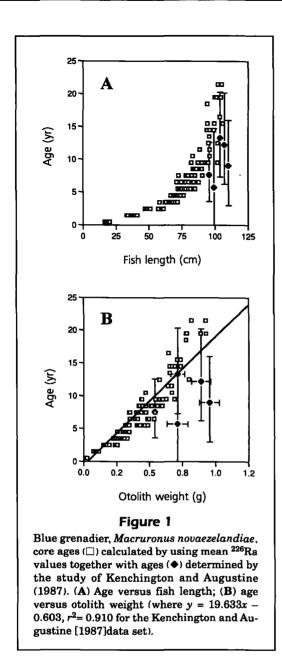
Otolith core ages were calculated by using both the individual sample 226 Ra value and the mean 226 Ra value. This has little effect on the average age of all the cores analyzed (e.g. 10.2 ± 5.0 years [with individual 226 Ra] and 9.6 ± 3.2 years [with mean 226 Ra]), but the maximum age recorded changes from 17.9 (+10.2, -7.7) years (with individual 226 Ra) to 13.3

(+7.1, -5.8) years (with mean ²²⁶Ra) depending on which radium value is used. Since all individual ²²⁶Ra activity ratios are similar within experimental error, it follows that the best age estimates are calculated by using the mean ²²⁶Ra activity.

Age estimates from annulus counts conducted by Kenchington and Augustine (1987) relative to fish length and otolith weight are plotted together with the radiometric core age estimates in Figure 1. The radiometric analysis of otolith cores are similar to those assigned by Kenchington and Augustine (1987), although there is some indication that the radiometric ages may be slightly lower. However, the core samples represent the average age of only three fish, the errors associated with the radiometric analyses are large, and limited sample sizes preclude any meaningful statistical analysis.

Discussion

Radiometric analysis of otolith cores has successfully provided age estimates for blue grenadier, in contrast to the unsuccessful analysis of whole otoliths by Fenton et al. (1990). The average age of the otolith core samples was approximately 10 years for fish 95—110 cm in length.



The 226 Ra concentrations in the blue grenadier otolith $(0.022\pm0.004~\mathrm{dpm\cdot g^{-1}})$ cores are low compared with other fish species. For example, orange roughy, Hoplostethus atlanticus, whole otoliths had a mean 226 Ra concentration 2–3 times higher (Fenton et al., 1991), and the otolith cores of three species of tropical snapper had 226 Ra values 6–10 times higher (Milton et al., 1995). The main consequence of the relatively low radium in blue grenadier cores is that the error associated with their age estimates are higher ± 4 –8 years than those for tropical snapper otolith cores ± 1 –2 years (Milton et al., 1995).

Furthermore, the ²²⁶Ra concentrations in the blue grenadier otolith cores were lower than the concen-

trations we reported earlier in whole otoliths from east coast 1+ fish $(0.050 \pm 0.005 \text{ to } 0.075 \pm 0.008 \text{ dpm} \cdot \text{g}^{-1})$ but higher than the values for 4+ fish $(0.016\pm 0.010 \text{ to } 0.018\pm 0.009 \text{ dpm}.\text{g}^{-1})$ (Fenton et al., 1990). The ²²⁶Ra concentrations found in Fenton et al. (1990) for whole blue grenadier otoliths ranged from $0.084 \pm 0.009 \text{ dpm} \cdot \text{g}^{-1}$ for 0+ fish caught in the Derwent estuary to $0.005 \pm 0.005 \text{ dpm} \cdot \text{g}^{-1}$ for 21+ fish caught off the west coast of Tasmania and exhibited a clear exponential reduction in ²²⁶Ra with fish length/age. The ²²⁶Ra found in cores in the present study are consistent with our earlier data for whole otoliths and also confirm that the core represents approximately a 2+ fish.

Reexamination of the radiometric data in our earlier study shows that data for a 1+ fish sampled from the east coast of Maria Island can also be used to estimate age. The sample had a 226 Ra value of 0.050 ± 0.005 dpm·g⁻¹ and 210 Pb of 0.003 ± 0.003 dpm·g⁻¹ which corresponds to a linear otolith mass growth radiometric age (see Fenton and Short, 1992, for the linear mass growth radiometric equation) of 0.68 ± 0.68 years. This is consistent with the age of 1+ determined by Kenchington and Augustine (1987). The remaining data in our earlier study cannot be used to determine age because of the exponential reduction in 226 Ra through life and the lack of a priori knowledge of the relationship between R and increasing otolith mass.

The age of Australian blue grenadier derived by radiometric age of otolith cores gives age estimates in approximate agreement with those derived by Kenchington and Augustine (1987) and New Zealand studies (reviewed Paul, 1992). Further radiometric ageing of otolith cores may be useful in examining apparent differences in the growth curves for New Zealand and Australian blue grenadier fisheries. For such a comparison, it would be worthwhile to include analysis with the radioisotope pair ²²⁸Th/²²⁸Ra because this should increase the precision of the age estimates particularly for fish under 5 years (Smith et al., 1991; Campana et al., 1993). In conclusion, the results of this study offer an independent assessment of age in blue grenadier and in doing so demonstrate the applicability of radiometric ageing of otolith cores for medium-aged temperate fish.

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